

# SEQUENCE LISTING

<110> Ajinomoto Co., Inc.

<120> A method of secreting and producing proteins

<130> Y1J0182

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<150> JP 2001-98808

<151> 2001-03-30

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<170> PatentIn Ver. 2.1

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 Asn Leu Lys Lys Glu Leu Ala Asn Gly Asn Asp Ala Leu Arg Asn Glu  
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 Lys Glu Arg Asn Gly Gly Asn His Asp Pro Ser Arg Met Lys Ala Val  
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 Ile Tyr Ser Lys His Phe Trp Ser Gly Gln Asp Arg Ser Ser Ser Ala  
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 His Tyr His Ala Pro Asn Gly Ser Leu Gly Ala Met His Val Tyr Glu  
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Val Pro Ala Asp Ala Ala Arg Leu Val Ala Ser Gly Lys Leu Asp Gln  
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Arg Leu Phe Asp Val Thr Glu Leu Asn Lys Ala Ala Thr Arg Thr Ala  
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His Arg Gly Gly Leu Lys Val Ile Val Gly Tyr Arg Gly Ala Ala Lys  
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Ala Ala Lys Ala Asp Val Arg Asp Ala Gly Thr Val Arg Arg Thr Leu  
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Thr Ser Leu Asn Ala Asp Ala Val Gln Thr Pro Gln Glu Ala Gly Ala  
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Ala Arg Val Trp Leu Asp Gly Val Arg Lys Ala Ser Leu Asp Thr Ser  
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Val Gly Gln Ile Gly Thr Pro Lys Ala Trp Glu Ala Gly Tyr Asp Gly  
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Pro Thr Thr Gly Asp Val Val Gly His Gly Thr His Val Ala Ser Ile  
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Pro Gly Ala Lys Ile Leu Asn Gly Lys Val Leu Asp Asp Ala Gly Phe  
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Lys Asp Lys Leu Ala Asp Phe Ser Ser Thr Gly Pro Arg Leu Gly Asp  
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Gln Trp Pro His Ala Asp Asp Lys Pro Val Thr Lys Lys Leu Thr Tyr  
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Arg Asn Leu Gly Thr Glu Asp Val Thr Leu Lys Leu Thr Ser Thr Ala  
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 Glu Thr Val Asn Thr Ala Val Phe Gly Pro Arg Leu Thr Ser Ser Tyr  
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Ala Leu Arg Ser Leu Leu Ala Ala Ser Met Leu Ile Gly Ala Ile Gly
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Glu Lys Pro Tyr Gln Gly Tyr Arg Tyr Leu Val Met Thr Tyr Arg Gln
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Pro Val Asp His Arg Asn Pro Gly Lys Gly Thr Phe Glu Gln Arg Phe
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Thr Leu Leu His Lys Asp Thr Asp Arg Pro Thr Val Phe Phe Thr Ser
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Gly Tyr Asn Val Ser Thr Asn Pro Ser Arg Ser Glu Pro Thr Arg Ile
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Arg Pro Gln Pro Ala Asp Trp Ser Lys Leu Asp Ile Trp Gln Ala Ala
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Phe Gln Asn Val Gly Asp Lys Ala Cys Arg Thr Gln Leu Asn Ser Val			
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Ala Gly Thr Gln Leu Gly Ala Pro Thr Val Lys Asn Pro His Leu Lys			
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ggc gtg ctg cgg tac ccc ggc atc aac cag ccg cgc tcg tac gtc ccc			1293
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Arg Asp Ile Pro Met Thr Phe Arg Pro Gly Ala Met Ala Asp Val Asp			
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cgc tgg gtg cgc gag gac agc cgg aac atg ctc ttc gtg tac ggg cag			1389
Arg Trp Val Arg Glu Asp Ser Arg Asn Met Leu Phe Val Tyr Gly Gln			
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Asn Asp Pro Trp Ser Gly Glu Pro Phe Arg Leu Gly Lys Gly Ala Ala			
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 35 40 45  
 Phe Val Glu Glu Lys Pro Tyr Gln Gly Tyr Arg Tyr Leu Val Met Thr  
 50 55 60  
 Tyr Arg Gln Pro Val Asp His Arg Asn Pro Gly Lys Gly Thr Phe Glu  
 65 70 75 80  
 Gln Arg Phe Thr Leu Leu His Lys Asp Thr Asp Arg Pro Thr Val Phe  
 85 90 95  
 Phe Thr Ser Gly Tyr Asn Val Ser Thr Asn Pro Ser Arg Ser Glu Pro  
 100 105 110  
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Ala Thr Tyr Phe Arg Arg Phe Tyr	Pro Asn Asp Met Asn Gly Thr Val	
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Ala Tyr Val Ala Pro Asn Asp	Val Asn Asp Lys Glu Asp Ser Ala Tyr	
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Asp Lys Phe Phe Gln Asn	Val Gly Asp Lys Ala Cys Arg Thr Gln Leu	
	210	215 220
Asn Ser Val Gln Arg Glu Ala Leu Val Arg Arg Asp Glu Ile Val Ala		
	225	230 235 240
Arg Tyr Glu Lys Trp Ala Lys Glu Asn Gly Lys Thr Phe Lys Val Val		
	245	250 255
Gly Ser Ala Asp Lys Ala Tyr Glu Asn Val Val Leu Asp Leu Val Trp		
	260	265 270
Ser Phe Trp Gln Tyr His Leu Gln Ser Asp Cys Ala Ser Val Pro Ala		
	275	280 285
Thr Lys Ala Ser Thr Asp Glu Leu Tyr Lys Phe Ile Asp Asp Ile Ser		
	290	295 300
Gly Phe Asp Gly Tyr Thr Asp Gln Gly Leu Glu Arg Phe Thr Pro Tyr		
	305	310 315 320
Tyr Tyr Gln Ala Gly Thr Gln Leu Gly Ala Pro Thr Val Lys Asn Pro		
	325	330 335
His Leu Lys Gly Val Leu Arg Tyr Pro Gly Ile Asn Gln Pro Arg Ser		
	340	345 350
Tyr Val Pro Arg Asp Ile Pro Met Thr Phe Arg Pro Gly Ala Met Ala		
	355	360 365
Asp Val Asp Arg Trp Val Arg Glu Asp Ser Arg Asn Met Leu Phe Val		
	370	375 380
Tyr Gly Gln Asn Asp Pro Trp Ser Gly Glu Pro Phe Arg Leu Gly Lys		
	385	390 395 400
Gly Ala Ala Ala Arg His Asp Tyr Arg Phe Tyr Ala Pro Gly Gly Asn		
	405	410 415
His Gly Ser Asn Ile Ala Gln Leu Val Ala Asp Glu Arg Ala Lys Ala		
	420	425 430
Thr Ala Glu Val Leu Lys Trp Ala Gly Val Ala Pro Gln Ala Val Gln		

435	440	445
Lys Asp Glu Lys Ala Ala Lys Pro Leu Ala Pro Phe Asp Ala Lys Leu		
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Asp Arg Val Lys Asn Asp Lys Gln Ser Ala Leu Arg Pro		
465	470	475

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<220>  
 <223> Description of Artificial Sequence:PCR primer

<400> 11  
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<210> 12  
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 <212> DNA  
 <213> Artificial Sequence

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 <223> Description of Artificial Sequence:PCR primer

<400> 12  
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<210> 13  
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 <212> DNA  
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 <223> Description of Artificial Sequence:PCR primer for  
 the promoter region and signal sequence region of  
 S.mobaraense

<400> 13  
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<210> 14  
 <211> 20  
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<220>  
 <223> Description of Artificial Sequence:PCR primer for  
 the promoter region and signal sequence region of  
 S.mobaraense

<400> 14  
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<210> 15  
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 <212> DNA  
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<220>  
 <223> Description of Artificial Sequence:PCR primer

<400> 15  
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<210> 16  
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 <212> DNA  
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<220>  
 <223> Description of Artificial Sequence:PCR primer

<400> 16  
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<210> 17  
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 <223> Description of Artificial Sequence:PCR primer

<400> 17  
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<210> 18  
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 <212> DNA  
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<220>  
 <223> Description of Artificial Sequence:PCR primer

<400> 18  
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<210> 19  
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<220>  
 <223> Description of Artificial Sequence:PCR primer  
  
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 <223> Description of Artificial Sequence:PCR primer  
  
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 <212> DNA  
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 <223> Description of Artificial Sequence:PCR primer  
  
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 <212> DNA  
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 <223> Description of Artificial Sequence:PCR primer  
  
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 <210> 27  
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 <212> DNA  
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 <223> Description of Artificial Sequence:PCR primer  
  
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 <210> 28  
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 <212> DNA  
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 <223> Description of Artificial Sequence:PCR primer  
  
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<220>  
<223> Description of Artificial Sequence:PCR primer

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19

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Arg Lys Trp Gln Gln Val Tyr Ser His Arg Asp Gly Lys Lys Gln Gln	
125 130 135	
atg acc gaa gag cag cga gaa aag ctg tcc tac ggt tgc gtt ggc gtc	606
Met Thr Glu Glu Gln Arg Glu Lys Leu Ser Tyr Gly Cys Val Gly Val	
140 145 150	
acc tgg gtc aac tcg ggc ccc tac ccg acg aac aga ttg gcg ttc gcg	654
Thr Trp Val Asn Ser Gly Pro Tyr Pro Thr Asn Arg Leu Ala Phe Ala	
155 160 165	
tcc ttc gac gag aac aag tac aag aac gac ctg aag aac acc agc ccc	702
Ser Phe Asp Glu Asn Lys Tyr Lys Asn Asp Leu Lys Asn Thr Ser Pro	
170 175 180	
cga ccc gat gaa acg cgg gcg gag ttc gag ggt cgc atc gcc aag ggc	750
Arg Pro Asp Glu Thr Arg Ala Glu Phe Glu Gly Arg Ile Ala Lys Gly	
185 190 195 200	
agt ttc gac gag ggg aag ggt ttc aag cgg gcg cgt gat gtg gcg tcc	798
Ser Phe Asp Glu Gly Lys Gly Phe Lys Arg Ala Arg Asp Val Ala Ser	
205 210 215	
gtc atg aac aag gcc ctg gaa aat gcc cac gac gag ggg act tac atc	846
Val Met Asn Lys Ala Leu Glu Asn Ala His Asp Glu Gly Thr Tyr Ile	
220 225 230	
aac aac ctc aag acg gag ctc acg aac aac aat gac gct ctg ctc cgc	894
Asn Asn Leu Lys Thr Glu Leu Thr Asn Asn Asn Asp Ala Leu Leu Arg	
235 240 245	
gag gac agc cgc tcg aac ttc tac tcg gcg ctg agg aac aca ccg tcc	942
Glu Asp Ser Arg Ser Asn Phe Tyr Ser Ala Leu Arg Asn Thr Pro Ser	
250 255 260	
ttc aag gaa agg gac ggc ggc aac tac gac ccg tcc aag atg aag gcg	990
Phe Lys Glu Arg Asp Gly Gly Asn Tyr Asp Pro Ser Lys Met Lys Ala	
265 270 275 280	
gtg atc tac tcg aag cac ttc tgg agc ggg cag gac cag cgg ggc tcc	1038
Val Ile Tyr Ser Lys His Phe Trp Ser Gly Gln Asp Gln Arg Gly Ser	
285 290 295	
tcc gac aag agg aag tac ggc gac ccg gaa gcc ttc cgc ccc gac cag	1086
Ser Asp Lys Arg Lys Tyr Gly Asp Pro Glu Ala Phe Arg Pro Asp Gln	
300 305 310	
ggt acc ggc ctg gtc gac atg tcg aag gac aga agc att ccg cgc agt	1134
Gly Thr Gly Leu Val Asp Met Ser Lys Asp Arg Ser Ile Pro Arg Ser	
315 320 325	
ccg gcc aag ccc ggc gaa ggt tgg gtc aat ttc gac tac ggt tgg ttc	1182
Pro Ala Lys Pro Gly Glu Gly Trp Val Asn Phe Asp Tyr Gly Trp Phe	

330                      335                      340  
 ggg gct caa aca gaa gcg gat gcc gac aaa acc aca tgg acc cac ggc 1230  
 Gly Ala Gln Thr Glu Ala Asp Ala Asp Lys Thr Thr Trp Thr His Gly  
 345                      350                      355                      360  
 gac cac tac cac gcg ccc aat agc gac ctg ggc ccc atg cac gta cac 1278  
 Asp His Tyr His Ala Pro Asn Ser Asp Leu Gly Pro Met His Val His  
 365                      370                      375  
 gag agc aag ttc cgg aag tgg tct gcc ggg tac gcg gac ttc gac cgc 1326  
 Glu Ser Lys Phe Arg Lys Trp Ser Ala Gly Tyr Ala Asp Phe Asp Arg  
 380                      385                      390  
 gga gcc tac gtg atc acg ttc ata ccc aag agc tgg aac acc gcc ccc 1374  
 Gly Ala Tyr Val Ile Thr Phe Ile Pro Lys Ser Trp Asn Thr Ala Pro  
 395                      400                      405  
 gcc aag gtg gag caa ggc tgg ccg tgacaggctg gtactacgac ctctgctgat 1428  
 Ala Lys Val Glu Gln Gly Trp Pro  
 410                      415  
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 <213> Streptoverdicillium cinnamoneum  
  
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 20                      25                      30  
 Gly Asp Gly Glu Glu Lys Gly Ser Tyr Ala Glu Thr His Gly Leu Thr  
 35                      40                      45  
 Ala Asp Asp Val Glu Ser Ile Asn Ala Leu Asn Glu Arg Ala Leu Thr  
 50                      55                      60  
 Leu Gly Gln Pro Gly Lys Pro Pro Lys Glu Leu Pro Pro Ser Ala Ser  
 65                      70                      75                      80  
 Ala Pro Ser Arg Ala Pro Ser Asp Asp Arg Glu Thr Pro Pro Ala Glu  
 85                      90                      95  
 Pro Leu Asp Arg Met Pro Glu Ala Tyr Arg Ala Tyr Gly Gly Arg Ala  
 100                      105                      110  
 Thr Thr Val Val Asn Asn Tyr Ile Arg Lys Trp Gln Gln Val Tyr Ser  
 115                      120                      125  
 His Arg Asp Gly Lys Lys Gln Gln Met Thr Glu Glu Gln Arg Glu Lys  
 130                      135                      140

Leu Ser Tyr Gly Cys Val Gly Val Thr Trp Val Asn Ser Gly Pro Tyr  
 145 150 155 160  
 Pro Thr Asn Arg Leu Ala Phe Ala Ser Phe Asp Glu Asn Lys Tyr Lys  
 165 170 175  
 Asn Asp Leu Lys Asn Thr Ser Pro Arg Pro Asp Glu Thr Arg Ala Glu  
 180 185 190  
 Phe Glu Gly Arg Ile Ala Lys Gly Ser Phe Asp Glu Gly Lys Gly Phe  
 195 200 205  
 Lys Arg Ala Arg Asp Val Ala Ser Val Met Asn Lys Ala Leu Glu Asn  
 210 215 220  
 Ala His Asp Glu Gly Thr Tyr Ile Asn Asn Leu Lys Thr Glu Leu Thr  
 225 230 235 240  
 Asn Asn Asn Asp Ala Leu Leu Arg Glu Asp Ser Arg Ser Asn Phe Tyr  
 245 250 255  
 Ser Ala Leu Arg Asn Thr Pro Ser Phe Lys Glu Arg Asp Gly Gly Asn  
 260 265 270  
 Tyr Asp Pro Ser Lys Met Lys Ala Val Ile Tyr Ser Lys His Phe Trp  
 275 280 285  
 Ser Gly Gln Asp Gln Arg Gly Ser Ser Asp Lys Arg Lys Tyr Gly Asp  
 290 295 300  
 Pro Glu Ala Phe Arg Pro Asp Gln Gly Thr Gly Leu Val Asp Met Ser  
 305 310 315 320  
 Lys Asp Arg Ser Ile Pro Arg Ser Pro Ala Lys Pro Gly Glu Gly Trp  
 325 330 335  
 Val Asn Phe Asp Tyr Gly Trp Phe Gly Ala Gln Thr Glu Ala Asp Ala  
 340 345 350  
 Asp Lys Thr Thr Trp Thr His Gly Asp His Tyr His Ala Pro Asn Ser  
 355 360 365  
 Asp Leu Gly Pro Met His Val His Glu Ser Lys Phe Arg Lys Trp Ser  
 370 375 380  
 Ala Gly Tyr Ala Asp Phe Asp Arg Gly Ala Tyr Val Ile Thr Phe Ile  
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 <212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:PCR primer

<400> 32

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21

<210> 33

<211> 32

<212> DNA

<213> Artificial Sequence

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<223> Description of Artificial Sequence:PCR primer

<400> 33

ggcggatcct cgcgtcgaga ggcgtggact ga

32

<210> 34

<211> 21

<212> DNA

<213> Artificial Sequence

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<223> Description of Artificial Sequence:PCR primer

<400> 34

tacgaattcg agctcggtac c

21

<210> 35

<211> 43

<212> DNA

<213> Artificial Sequence

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<223> Description of Artificial Sequence:PCR primer

<400> 35

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43

<210> 36

<211> 26

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:PCR primer

<400> 36

aacggggaga acagcacggc cgccgg

26

<210> 37  
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 <223> Description of Artificial Sequence:PCR primer  
  
 <400> 37  
 ggcgaattct ccggcgggcc gtcaccggt 29

<210> 38  
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 <220>  
 <223> Description of Artificial Sequence:PCR primer for  
 fused prepro-serineprotease construction  
  
 <400> 38  
 ggcaagctta aattcctgtg aattagctga 30

<210> 39  
 <211> 44  
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<210> 40  
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 <212> PRT  
 <213> Streptovercillium mobaraence  
  
 <400> 40  
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 Val Glu Glu Lys  
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<210> 41  
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 <212> PRT  
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<223> Description of Artificial Sequence:probe for svPEP

<400> 41

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<210> 42

<211> 33

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:probe for svPEP

<400> 42

aagatccccg ggatgaagtt cgatcgaggag aag

33

<210> 43

<211> 23

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:PCR primer

<400> 43

gaggcggcgt cgatcaccgc ccc

23

<210> 44

<211> 32

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:PCR primer

<400> 44

gccaagcttg aagcaccggc ggcggcaccg gg

32

<210> 45

<211> 46

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:PCR primer

<400> 45

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46

<210> 46

<211> 37

<212> DNA  
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 <210> 47  
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 <210> 48  
 <211> 42  
 <212> DNA  
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 <223> Description of Artificial Sequence:PCR primer  
  
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 <400> 49  
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 <210> 50  
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 <400> 50  
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<210> 51  
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<220>  
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<400> 51  
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<210> 52  
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<212> DNA  
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<400> 52  
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<210> 53  
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<400> 53  
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<210> 54  
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<220>  
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<400> 54  
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<210> 55  
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<212> PRT  
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<400> 55  
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Cys Val Val Gly Tyr Ile Gly Glu Arg Cys Gln Tyr Arg Asp Leu Lys  
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Trp Trp Glu Leu Arg  
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<210> 56  
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 <212> DNA  
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<220>  
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<210> 57  
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 <212> DNA  
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<220>  
 <223> Description of Artificial Sequence:PCR primer

<400> 57  
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<210> 58  
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 <212> DNA  
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<210> 59  
 <211> 20  
 <212> DNA  
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<220>  
 <223> Description of Artificial Sequence:PCR primer

<400> 59  
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<210> 60  
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<213> Streptovercillium mobaraense

<400> 60  
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1